

FIGURE 1.

CONSENSUS SEQUENCE OF MC4R IN PIG

!!NA_SEQUENCE 1.0
 con-mc4r.seq Length: 746 April 9, 1998 16:17 Type: N Check: 125

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1  ACAAGAAATCT GCATTACCC ATGTACTTTT TCATCTGTAG CCTGGCTGTG
51  GCTGATATGC TGGTGAGCGT TTCCAATGGG TCAGAAACCA TTGTCATCAC
101 CCTATTAAAC AGCACGGACA CGGACGCACA GAGTTTCACA GTGAATATTG
151 ATAATGTCAT TGA CTGAGTG ATCTGTAGCT CCTTACTCGC CTC AATTTGC
201 AGCCTGCTTT CGATTGCAGT GGACAGGTAT TTTACTATCT TTTATGCTCT
251 CCAGTACCAT AACATTATGA CAGTTAAGCG GGTGGAATC ATCATCAGTT
301 GTATCTGGGC AGTCTGCACG GTGTCGGGTG TTTTGTTCAT CATTACTCA
351 GATAGCAGTG CTGTTATTAT CTGCCTCATA ACCGTGTTCT TCACCATGCT
401 GGTCTCATG GCTTCTCTCT ATGTCCACAT GTTCCTCATG GCCAGACTCC
451 ACATTAAGAG GATCGCCGTC CTCCCAGGCA CTGGCACCAT CCGCCAAGGT
501 GCCAACATGA AGGGGGCAAT TACCCTGACC ATCTTGATTG GGGTCTTTGT
551 GGTCTGCTGG GCCCCCTTCT TCCTCCACTT AATATTCTAT ATCTCCTGCC
601 CCCAGAATCC ATACTGTGTG TGCTTCATGT CTCACTTTAA TTTGTATCTC
651 ATCCTGATCA TGTGTAATTC CATCATCAT CCCCTGATTT ATGCACTCCG
701 GAGCCAAGAA CTGAGGAAAA CCTTCAAAGA GATCATCTGT TGCTAT

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x = G or A, the site of polymorphism.

FIGURE 2.

COMPARISON OF DNA SEQUENCE BETWEEN HUMAN AND PORCINE *MC4R* GENE

(Nucleotide) FASTA of: con-mc4r.seq from: 1 to: 746 April 10, 1998 19:52

TO: s77415.gb_pr Sequences: 1 Symbols: 1,671 Word Size: 6

Searching with both strands of the query.

Scoring matrix: GenRunData:fastadna.cmp

Constant pamfactor used

Gap creation penalty: 16 Gap extension penalty: 4

The best scores are: init1 initn opt..

/usr2/users/rothschi/kwan/mc4r/seq/s77415.gb_pr Begin: 608 End: 1353

! LOCUS S77415 1671 bp ... 3208 3208 3208

/usr2/users/rothschi/kwan/mc4r/seq/s77415.gb_pr Begin: 388 End: 407

Strand: -

! LOCUS S77415 1671 bp ... 70 70 73

\\End of List

con-mc4r.seq

/usr2/users/rothschi/kwan/mc4r/seq/s77415.gb_pr

LOCUS S77415 1671 bp DNA PRI 26-SEP-1995

DEFINITION melanocortin-4 receptor (human, Genomic, 1671 nt).

ACCESSION S77415

NID g998456

KEYWORDS

SOURCE human. . . .

SCORES Init1: 3208 Initn: 3208 Opt: 3208

92.2% identity in 746 bp overlap

(SEQU ID NO:3)

con-mc4r.seq

10 20 30
ACNAGATCTGCATTACCCGATGACTTTT
|||||

(SEQU ID NO:2)

s77415

ATATCTTAGTGATTGTGCAATAGCCAGAACAGAAATCTGCATTACCCGATGACTTTT
580 590 600 610 620 630

40 50 60 70 80 90
con-mc4r.seq TCATCTGTAGCTGGCTGTGGCTGATATGCTGGTGAGCGTTTCCTCAATGGGTGAGAACCA
|||||
s77415 TCATCTGCAGCTTGGCTGTGGCTGATATGCTGGTGAGCGTTTCCTCAATGGGTGAGAACCA
640 650 660 670 680 690

100 110 120 130 140 150
con-mc4r.seq TTGTCATCACCCCTATTAAACAGCACGGACACGGACGCACAGAGTTTCACAGTGAAATATTG
|||||
s77415 TTATCATCACCCCTATTAAACAGTACAGATACGGATACAGAGTTTCACAGTGAAATATTG
700 710 720 730 740 750

FIGURE 2. (cont.)

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con-mc4r.seq 160      170      180      190      200      210
ATAATGTCATTGACTCAGTGATCTGTAGCTCCTTACTCGGCTCAATTTGCAGCCTGCTTT
s77415      160      170      180      190      200      210
ATAATGTCATTGACTCGGTGATCTGTAGCTCCTTGTGCTTCATCCATTTGCAGCCTGCTTT
760      770      780      790      800      810

con-mc4r.seq 220      230      240      250      260      270
CGATTGTCAGTGGACAGGTATTTTACTATCTTTTATGCTCTCCAGTACCATAAGCATATGA
s77415      220      230      240      250      260      270
CAATTGTCAGTGGACAGGTACTTTTACTATCTTTTATGCTCTCCAGTACCATAAGCATATGA
820      830      840      850      860      870

con-mc4r.seq 280      290      300      310      320      330
CAGTTTAAGCGGGTTGGATCATCATCAGTTGTATCTGCGCAGTCTGCACGGTGTGCGGGTG
s77415      280      290      300      310      320      330
CAGTTTAAGCGGGTTGGATCAGCATTAAGTTGTATCTGCGCAGTCTGCACGGTGTTCAGGCA
880      890      900      910      920      930

con-mc4r.seq 340      350      360      370      380      390
TTTTGTTTCATTCTACTCAGATAGCAGTGCTGTTATTATCTGCCATCAACCGTGTCTT
s77415      340      350      360      370      380      390
TTTTGTTTCATTCTACTCAGATAGTAGTGCTGTCATCATCTGCCCTCATCACCATTGTTCT
940      950      960      970      980      990

con-mc4r.seq 400      410      420      430      440      450
TCACCATGCTGGCTCTCATGGCTTCTCTCTATGTCACATGTTCTTCATGGCCAGACTCC
s77415      400      410      420      430      440      450
TCACCATGCTGGCTCTCATGGCTTCTCTCTATGTCACATGTTCTTCATGGCCAGGCTTC
1000     1010     1020     1030     1040     1050

con-mc4r.seq 460      470      480      490      500      510
ACATTAAAGAGGATCGCGCTCTCCCGAGGCACCTGGCACCATCCGCCAAGGTGCCAACATGA
s77415      460      470      480      490      500      510
ACATTAAAGAGGATGCTGCTCTCCCGAGGCACCTGGTGCACCATCCGCCAAGGTGCCAACATGA
1060     1070     1080     1090     1100     1110

con-mc4r.seq 520      530      540      550      560      570
AGGGGGCAATTACCCTGACCATCTTGATTGGGGTCTTTTGTGGTCTGCTGGGGCCCCCTCT
s77415      520      530      540      550      560      570
AGGGAGCGATTACCTTGACCATCTTGATTGGGGTCTTTTGTGGTCTGCTGGGGCCCCCATCT
1120     1130     1140     1150     1160     1170

con-mc4r.seq 580      590      600      610      620      630
TCCCTCACTTAATATTCTATATCTCTGCCCCCAGAATCCATCTGTGTGTGCTTCATGT
s77415      580      590      600      610      620      630
TCCCTCACTTAATATTCTATATCTCTGCCCCCAGAATCCATCTGTGTGTGCTTCATGT
1180     1190     1200     1210     1220     1230

con-mc4r.seq 640      650      660      670      680      690
CTCACCTTAATTTGTATCTCATCTGATCATGTGTAATTCATCATCAATCCCTCGATTT
s77415      640      650      660      670      680      690
CTCACCTTAATTTGTATCTCATCTGATCATGTGTAATTCATCATCATCTCTCTGATTT
1240     1250     1260     1270     1280     1290

con-mc4r.seq 700      710      720      730      740
ATGCACCTCCGGAGCCAAAGACTGAGGAAACCTTCAAGAGATCATCTGTTGCTAT
s77415      700      710      720      730      740
ATGCACCTCCGGAGCTCAAGACTGAGGAAACCTTCAAGAGATCATCTGTTGCTATCCCC
1300     1310     1320     1330     1340     1350

s77415      1360     1370     1380     1390     1400     1410
TGGGAGGCCCTTTGTGACTTGTCTAGCAGATATTAATGGGACAGAGCACGCAATATAGG

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FIGURE 3.

COMPARISON OF AMINO ACID SEQUENCE BETWEEN HUMAN AND
PORCINE MC4R GENE

(Peptide) TFASTA of: human.pep from: 1 to: 332 April 9, 1998 16:18

REFORMAT of: human check: 9754 from: 1 to: 332 March 7, 1998 18:43
(No documentation)

TO: mc4r-allele.seq Sequences: 2 Symbols: 1,492 Word Size: 2

Searching all six frames.

Scoring matrix: GenRunData:blosum50.cmp

Variable pamfactor used

Gap creation penalty: 16 Gap extension penalty: 4

The best scores are: frame init1 initn opt..

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/usr2/users/rothschi/kwan/mc4r/seq/mc4r-allele1.seq Begin: 3 End: 746
! mc4r-allele1.seq Length: 746 April ... (3) 1602 1602 1602
/usr2/users/rothschi/kwan/mc4r/seq/mc4r-allele2.seq Begin: 3 End: 746
! mc4r-allele2.seq Length: 746 April ... (3) 1596 1596 1596

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SCORES Frame: (3) Init1: 1602 Initn: 1602 Opt: 1602
97.6% identity in 248 aa overlap

(Seq ID NO: 4)

	50	60	70	80	90	100
human.pep	QLFVSPFEVFTLGVISLLENILVIVAIKNNKLNHSPMYFFICSLAVADMLVSVNGSETI					
mc4r-allele1				KLNHSPMYFFICSLAVADMLVSVNGSETI		
				10	20	30

(Seq ID NO: 5)

	110	120	130	140	150	160
human.pep	IITLLNSTDTPAQSFVNIDNVIDSVICSSLLASICSLLSIAVDRYFTIFYALQYHNIMT					
mc4r-allele1	VITLLNSTDTPAQSFVNIDNVIDSVICSSLLASICSLLSIAVDRYFTIFYALQYHNIMT					
	40	50	60	70	80	90

	170	180	190	200	210	220
human.pep	VKRVGISISCIWAQCTVSGILFIIYSDSSAVIICLITMFFTMLALMASLYVHMFILMARLH					
mc4r-allele1	VKRVGIIISCIWAVCTVSGVLFIIYSDSSAVIICLITVFFTMLALMASLYVHMFILMARLH					
	100	110	120	130	140	150

	230	240	250	260	270	280
human.pep	IKRIAVLPGTGAIROGANNKGAITLTILIGVVFVVCWAPFFLHLIFYISCPQNPYCVCFMS					
mc4r-allele1	IKRIAVLPGTGTIROGANNKGAITLTILIGVVFVVCWAPFFLHLIFYISCPQNPYCVCFMS					
	160	170	180	190	200	210

	290	300	310	320	330
human.pep	HFNLYLILIMCNSIIDPLIYALRSQELRKTFFKEIICCYPLGGCLDSLSSRY				
mc4r-allele1	HFNLYLILIMCNSIIDPLIYALRSQELRKTFFKEIICCY				
	220	230	240		

human.pep
/usr2/users/rothschi/kwan/mc4r/seq/mc4r-alle2.seq

SCORES Frame: (3) Init1: 1596 Initn: 1596 Opt: 1596
97.2% identity in 248 aa overlap

	50	60	70	80	90	100
human.pep	QLFVSPEVFTLGVISLLENILVTVAIAKKNLHSPMYFFICSLAVADMLVSVSNGSETI					
mc4r-alle2				KNLHSPMYFFICSLAVADMLVSVSNGSETI		
				10	20	30
	110	120	130	140	150	160
human.pep	IITLLNSTDTDAQSFTVNIDNVIDSVICSSLLASICSLLSIAVDRYFTIFYALQYHNIMT					
mc4r-alle2	VITLLNSTDTDAQSFTVNIDNVIDSVICSSLLASICSLLSIAVDRYFTIFYALQYHNIMT					
	40	50	60	70	80	90
	170	180	190	200	210	220
human.pep	VKRVGISISCIWAACVSGILFIIYSDSSAVIICLITMFFTMLALMASLYVHMFILMARLH					
mc4r-alle2	VKRVGIIISCIWAVCTVSGVLFIIYSDSSAVIICLITVFFTMLALMASLYVHMFILMARLH					
	100	110	120	130	140	150
	230	240	250	260	270	280
human.pep	IKRIAVLPGTGAIROGANMKGAITLTILIGVFVVCWAPFFLHLIFYISCPQNPYCVCFMS					
mc4r-alle2	IKRIAVLPGTGAIROGANMKGAITLTILIGVFVVCWAPFFLHLIFYISCPQNPYCVCFMS					
	160	170	180	190	200	210
	290	300	310	320	330	
human.pep	HFNLYLILIMCNSIIDFLIYALRSQELRKTfKEIICCYPLGGCLDLSRY					
mc4r-alle2	HFNLYLILIMCNSIINPLIYALRSQELRKTfKEIICCY					
	220	230	240			

! CPU time used:
! Database scan: 0:00:00.2
! Post-scan processing: 0:00:00.1
! Total CPU time: 0:00:00.4
! Output File: human.tfasta

LINKAGE REPORT OF TWO-POINT ANALYSIS

New locus MC4R
in sex-equal linkage analyses with existing loci

S0082	MC4R	rec. fracs. =	0.05,	lods =	14.74
CGA	MC4R	rec. fracs. =	0.14,	lods =	6.88
S0020	MC4R	rec. fracs. =	0.18,	lods =	5.32
S0079	MC4R	rec. fracs. =	0.12,	lods =	10.35
S0155	MC4R	rec. fracs. =	0.14,	lods =	7.68
S0122	MC4R	rec. fracs. =	0.18,	lods =	5.17
S0313	MC4R	rec. fracs. =	0.00,	lods =	17.76
S0312	MC4R	rec. fracs. =	0.20,	lods =	5.60
S0311	MC4R	rec. fracs. =	0.17,	lods =	7.18
S0416	MC4R	rec. fracs. =	0.20,	lods =	3.21
S0331	MC4R	rec. fracs. =	0.02,	lods =	21.91
S0396	MC4R	rec. fracs. =	0.16,	lods =	7.85
BHT0433	MC4R	rec. fracs. =	0.02,	lods =	21.32
S0536	MC4R	rec. fracs. =	0.03,	lods =	15.61
CAPN3	MC4R	rec. fracs. =	0.12,	lods =	6.65
KGF	MC4R	rec. fracs. =	0.09,	lods =	6.46
MEF2A	MC4R	rec. fracs. =	0.05,	lods =	14.36
MC4R	MC4R	rec. fracs. =	0.00,	lods =	26.19

new locus MC4R

in sex-specific (female/male) linkage analyses with existing loci

S0082	MC4R	rec. fracs.=	0.00	0.09,	lods =	15.86
CGA	MC4R	rec. fracs.=	0.07	0.22,	lods =	7.46
S0020	MC4R	rec. fracs.=	0.00	0.25,	lods =	6.33
S0079	MC4R	rec. fracs.=	0.00	0.19,	lods =	11.48
S0155	MC4R	rec. fracs.=	0.00	0.24,	lods =	9.98
S0122	MC4R	rec. fracs.=	0.00	0.27,	lods =	7.10
S0313	MC4R	rec. fracs.=	0.00	0.00,	lods =	17.76
S0312	MC4R	rec. fracs.=	0.04	0.29,	lods =	7.45
S0311	MC4R	rec. fracs.=	0.00	0.28,	lods =	9.02
S0416	MC4R	rec. fracs.=	0.00	0.31,	lods =	4.17
S0331	MC4R	rec. fracs.=	0.05	0.00,	lods =	22.14
S0396	MC4R	rec. fracs.=	0.03	0.24,	lods =	9.33
BHT0385	MC4R	rec. fracs.=	0.14	0.36,	lods =	3.46
BHT0433	MC4R	rec. fracs.=	0.05	0.00,	lods =	21.82
S0536	MC4R	rec. fracs.=	0.00	0.05,	lods =	15.77
CAPN3	MC4R	rec. fracs.=	0.00	0.18,	lods =	7.35
KGF	MC4R	rec. fracs.=	0.00	0.17,	lods =	6.74
MEF2A	MC4R	rec. fracs.=	0.10	0.00,	lods =	14.52
MC4R	MC4R	rec. fracs.=	0.00	0.00,	lods =	26.19

LINKAGE REPORT OF MULTIPOINT ANALYSIS

Sex_averaged map (recomb. frac., Kosambi cM):

0	ESR			0.0
		0.18	18.4	
1	S0008			18.4
		0.12	11.9	
7	CGA			30.3
		0.03	2.8	
3	S0312			33.1
		0.05	4.9	
4	S0122			38.1
		0.09	9.4	
8	KGF			47.4
		0.06	5.8	
6	CAPN3			53.2
		0.02	2.5	
9	MEF2A			55.7
		0.06	6.1	
5	MC4R			61.8
		0.06	5.6	
10	S0313			67.4
		0.00	0.0	
11	S0082			67.4
		0.03	3.4	
12	S0079			70.8
		0.03	2.5	
14	S0142			73.3
		0.01	1.0	
13	S0020			74.4
		0.04	4.3	
15	S0311			78.7
		0.00	0.0	
16	S0155			78.7
		0.12	12.2	
17	S0113			90.9
		0.20	21.0	
18	S0302			111.9
		0.22	23.4	
19	S0112			135.3

* denotes recomb. frac. held fixed in this analysis. log10_like = -305.098

SESS ID NO: 12

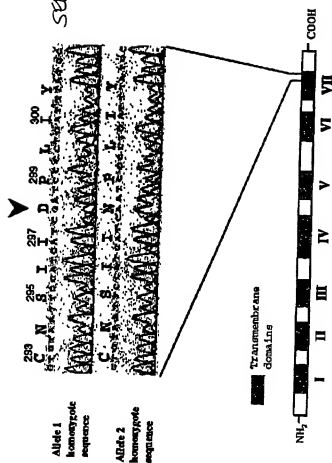
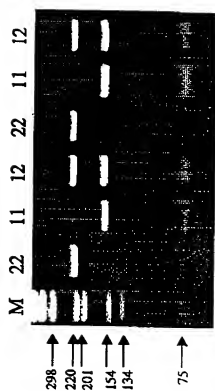


FIGURE 6.



Sequence alignment of various cannabinoid receptors. The alignment shows amino acid sequences for pMC4R, hMC4R, rMC4R, sheep MC5R, bovine MC5R, bovine MC2R, hMC3R, mMC3R, hMC2R, hMC1R, bEDG-2R, hEDG-4R, human cannab, hH2AB, hSSP2, and hGAL1-R. The sequences are aligned horizontally, with positions 1 to 306 indicated at the top. A large arrow on the left points to the start of the alignment.